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## Trypsin Digestion of Formalin-Fixed, Paraffin-Embedded Tissues Prior to Immunohistochemistry

1. Preheat the following to 37 °C using a water bath:
  - (i) 200ml of TBS
  - (ii) 200ml of distilled water.
2. Dissolve 0.2g Trypsin 250 and 0.2g Calcium Chloride in the 200ml of TBS.
3. Once the trypsin solution is at 37 °C, pH to 7.8 with 1M sodium hydroxide.
4. Place rehydrated paraffin sections in the distilled water to preheat the sections to 37 °C for a minimum of 5 minutes.
5. Incubate sections in trypsin solution at 37 °C. The time required will depend on the antibody and tissue, however, 30 minutes is usually sufficient.
6. Rinse sections in running tap water.
7. Proceed with immunohistochemistry protocol.

### **Equipment and Materials Required:**

50mM Tris-buffered saline

Trypsin 250: Difco order code 0152-13 (available from Becton Dickinson).

Calcium chloride

1M Sodium Hydroxide

**NB. Trypsin containing chymotrypsin should always be used. The enzyme activities can vary from a supplier and between batches. Such variations may affect the incubation time required.**